

**METHOD DEVELOPMENT AND ANALYSIS OF
CYPERMETHRIN AND λ -CYHALOTHRIN IN PALM OIL
MATRICES AND SOIL SAMPLES BY GAS
CHROMATOGRAPHY**

BADRUL HISYAM ZAINUDIN

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**A DISSERTATION SUBMITTED TO THE FACULTY OF
SCIENCE UNIVERSITY OF MALAYA IN FULFILMENT OF
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ABSTRACT

In this study, new sample preparation methods were developed for the determination of cypermethrin and λ -cyhalothrin residues in crude palm oil (CPO), crude palm kernel oil (CPKO), and soil samples using gas chromatography with electron capture detector (GC-ECD). The first half of the thesis studied the development and optimization of pesticides extraction from palm oil samples. Sample preparations were divided into two parts. The first part was the extraction step based on low-temperature extraction technique. Factors such as acetonitrile volume, decanting the liquid phase from the precipitated oil after low-temperature precipitation, and the freezing time were studied and optimized. In the second part, two different clean-up methodologies were tested for co-extractives elimination: (i) solid-phase extraction (SPE); and (ii) dispersive solid-phase extraction (d-SPE). In both techniques, different sorbents were studied and optimized. Finally, the optimized conditions were obtained using acetonitrile extraction followed by low-temperature precipitation. Then, oil extracts were cleaned-up using combined SPE employing graphitized carbon black (GCB) and primary secondary amine (PSA) as the optimized sorbents.

The second half of the thesis studied the development and optimization of pesticides extraction from soil samples. Ultrasonic extraction (USE) was chosen as the extraction technique for both cypermethrin and λ -cyhalothrin. Extraction parameters such as extraction solvent (acetonitrile, ethyl acetate, and acetone), solvent volume (15, 20, and 25 mL), and duration of sonication (5, 10, 15, and 20 min) were comprehensively investigated and optimized. The optimized conditions were obtained using 15 mL of acetonitrile as the extraction solvent and 5 min as the duration of sonication.

Both developed methods gave good linearity, detection limits, precision and sensitivity for cypermethrin and λ -cyhalothrin. Good regression coefficients (>0.99) were obtained in all cases in the linearity study. The detection limits were 0.025 and 0.05 $\mu\text{g/g}$ for λ -cyhalothrin and cypermethrin respectively for palm oil matrices. On the other hand, for soil samples, the detection limits were 0.0025 and 0.01 $\mu\text{g/g}$ for λ -cyhalothrin and cypermethrin respectively. Furthermore, the optimized method also gave acceptable recoveries ($> 80\%$) and good precision in terms of RSDs ($< 7\%$). Finally, the newly proposed sample preparation techniques for cypermethrin and λ -cyhalothrin were applied to crude palm oil samples obtained from refineries and real soil samples obtained from oil palm plantation. The results suggested that both field samples were free from contamination of pesticides of interest.

ABSTRAK

Dalam kajian ini, kaedah penyediaan sampel yang baru telah dibangunkan bagi menentukan sisa baki cypermethrin dan λ -cyhalothrin dalam minyak sawit mentah, minyak isirung sawit mentah, dan sampel tanah menggunakan kromatografi gas dengan pengesan tangkapan elektron (GC-ECD). Bahagian pertama tesis ini mengkaji pembangunan dan pengoptimuman bagi pengekstrakan racun perosak daripada sampel minyak sawit. Penyediaan sampel telah dibahagikan kepada dua bahagian. Bahagian yang pertama adalah langkah pengekstrakan yang berasaskan teknik pengekstrakan suhu rendah. Faktor-faktor seperti isipadu asetonitril, penyirangan fasa cecair daripada minyak yang termendak selepas pemendakan suhu rendah, dan masa pembekuan telah dikaji dan dioptimumkan. Dalam bahagian yang kedua, dua kaedah pembersihan yang berlainan telah diuji untuk menyingkirkan ko-pengekstrakan: (i) pengekstrakan fasa pepejal (SPE); (ii) pengekstrakan fasa pepejal menyelerak (d-SPE). Dalam kedua-dua teknik, penjerap yang berlainan telah dikaji dan dioptimumkan. Akhir sekali, keadaan yang paling optimum diperolehi dengan pengekstrakan menggunakan asetonitril diikuti dengan pemendakan suhu rendah. Kemudian, ekstrak bagi minyak dibersihkan dengan gabungan dua SPE menggunakan karbon grafit hitam (GCB) dan amina primer sekunder (PSA) yang merupakan penjerap paling optimum.

Bahagian kedua tesis mengkaji pembangunan dan pengoptimuman pengekstrakan racun perosak daripada sampel tanah. Pengekstrakan secara ultrasonik telah dipilih sebagai teknik pengekstrakan bagi kedua-dua cypermethrin dan λ -cyhalothrin. Parameter pengekstrakan seperti pelarut ekstrak (asetonitril, etil asetat, dan aseton), isipadu pelarut (15, 20, dan 25 mL), dan masa sonifikasi (5, 10, 15, dan 20 min) telah dikaji dengan terperinci dan dioptimumkan. Keadaan yang paling optimum adalah

diperolehi dengan menggunakan 15 mL asetonitril sebagai pelarut pengekstrakan dan masa sonifikasi selama 5 minit.

Kedua-dua kaedah yang dibangunkan memberikan kelinearan, had pengesanan, kepersisan, dan kepekaan yang baik untuk cypermethrin dan λ -cyhalothrin. Pemalar linear yang baik (>0.99) telah diperolehi bagi semua kes dalam kajian kelinearan. Had pengesanan adalah 0.025 dan 0.05 $\mu\text{g/g}$ masing-masing bagi λ -cyhalothrin dan cypermethrin untuk matriks minyak sawit. Manakala, bagi sampel tanah, had pengesanan adalah 0.0025 dan 0.01 $\mu\text{g/g}$ masing-masing untuk λ -cyhalothrin dan cypermethrin. Selain itu, kaedah yang optimum juga memberikan perolehan kembali yang boleh diterima ($> 80\%$) dan kepersisan yang baik dalam bentuk nilai RSD ($< 7\%$). Akhir sekali, teknik penyediaan sampel yang dicadangkan untuk cypermethrin dan λ -cyhalothrin telah digunakan kepada sampel minyak sawit mentah yang diperolehi dari kilang penapisan dan sampel tanah yang diperolehi dari ladang kelapa sawit. Keputusan yang diperolehi menunjukkan kedua-dua sampel berkenaan adalah bebas daripada pencemaran racun perosak yang dikaji.

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LIST OF ABBREVIATIONS

ACN	- Acetonitrile
ANOVA	- Analysis of variance
ASE	- Accelerated solvent extraction
CBE	- Cocoa butter equivalent
CPKO	- Crude palm kernel oil
CPO	- Crude palm oil
CV	- Coefficient of variation
DAD	- Diode array detector
DPV	- Differential pulse voltammetry
DPX	- Disposable pipette extraction
d-SPE	- Dispersive solid-phase extraction
ECD	- Electron capture detector
FAO	- Food and Agriculture Organization
FD	- Fluorescence
FFB	- Fresh fruit bunch
FID	- Flame ionization detector
FPD	- Flame photometric detector
GAP	- Good agricultural practice
GC	- Gas chromatography
GCB	- Graphitized carbon black
GC-ECD	- Gas chromatography-electron capture detector
GPC	- Gel permeation chromatography
HLLE	- Homogenous liquid-liquid extraction
HPLC	- High performance liquid chromatography

hr	- Hours
HS-SPME	- Headspace solid-phase microextraction
ICI	- Imperial Chemical Industry
IUPAC	- International Union of Pure and Applied Chemistry
$K_{o/w}$	- Octanol-water partition coefficient
LC	- Liquid chromatography
LOD	- Limit of detection
LOQ	- Limit of quantification
LPME	- Liquid-phase microextraction
Min	- Minutes
MPOB	- Malaysian Palm Oil Board
MRL	- Maximum residue limit
MS	- Mass spectrometric
MSPD	- Matrix solid-phase dispersion
MTOAC	- Methyltrioctylammonium chloride
NPD	- Nitrogen-phosphorus detector
OCP	- Organochlorine pesticide
OPP	- Organophosphate pesticide
PAHs	- Polyaromatic hydrocarbons
PCBs	- Polychlorinated biphenyls
PLE	- Pressurised liquid extraction
PSA	- Primary secondary amine
QuEChERS	- Quick Easy Cheap Efficient Rugged Safe
RBDPO	- Refined bleached deodorized palm oil
RSD	- Relative standard deviation
S/N	- Signal-to-noise

SCD	- Sweep co-distillation
SFE	- Supercritical fluid extraction
SPE	- Solid-phase extraction
SPME	- solid-phase microextraction
TCD	- Thermal conductivity cell detector
USE	- Ultrasonic extraction
UV	- Ultraviolet
WCOT	- Wall coated open tubular